

Chilina megastoma Hylton Scott, 1958 (Pulmonata: Basommatophora): a study on topotypic specimens

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Abstract: This is a contribution to the knowledge of *Chilina megastoma* Hylton Scott, 1958 (Pulmonata: Basommatophora: Chiliniidae), based on the study of topotypic specimens. This species is endemic to the Iguazú Falls, a frontier region between Argentina and Brazil. *C. megastoma* is known only from a brief original description, which was based exclusively on the shell morphology of a single specimen. The relevant results include the following: the radula is formed by 40-50 transverse rows of teeth in "chevron" arrangement; the central tooth is bicuspid with a reduced or rudimentary third cusp; the first lateral is tricuspid with a wide and short base, and the remaining laterals and marginals (40-44) each have four cusps and a long attachment base, which is bent in the marginals. The secondary or accessory seminal receptacle, in the female genital system, is strongly reduced and does not seem to be functional. The penis is as long as the penis sheath, and the penis sheath is two or three times longer than the prepuce.

Key words: *Chilina*, morphology, topotypes, endemic species, Chiliniidae

The family Chiliniidae comprises only one genus, *Chilina* Gray, 1828, which is distributed exclusively in South America (Castellanos and Gaillard, 1981; Castellanos and Miquel, 1991). About 17 *Chilina* species have been recognized as distributed in Argentina (Castellanos and Gaillard, 1981; Castellanos and Landoni, 1995).

Previous knowledge on the soft-part anatomy of members of the genus mainly comes from the studies of Haeckel (1911) and Duncan (1960a, b; 1975). Recently, Valdovinos and Stuardo (1995), based upon study of the radula, soft-part anatomy, and shell morphology of the 36 species of the genus described from Chile, proposed a new genus, *Archaeochilina*, to include *Chilina angusta* (Philippi, 1860), and recommended the rearrangement of the remaining Chilean species in three new subgenera: *Chilina* s. s., *Mesochilina*, and *Neochilina*. The authors proposed the nervous system organization as providing the only reliable character to afford a basis for supraspecific arrangement.

Argentine species of *Chilina* (16 species with three subspecies) have been studied by Hylton Scott (1958), Castellanos and Gaillard (1981), Castellanos and Miquel (1980), and Miquel (1984; 1986a, b).

Chilina megastoma Hylton Scott, 1958, was described from the Iguazú Falls (25°35' S, 54°35' W), Misiones Province, Argentina. The species is known only

from the type locality (Castellanos and Landoni, 1995). The original description was based on the shell morphology of a single specimen, thus, details on soft-part anatomy, especially the nervous system, genital system, and radular features, are completely unknown.

In the present paper, the results of the study on topotypic specimens of *Chilina megastoma*, focusing on its shell morphology, radular ultrastructure, anatomy of the male and female genital systems, and nervous system, are given. In addition, *C. fluminea* (Maton, 1809), a species widely distributed in Argentina, was used for comparative analysis of several anatomical features.

MATERIALS AND METHODS

Two samples of *Chilina megastoma* were obtained, one from the vertical cliff behind the Salto Dos Hermanas waterfall on the Argentine side of the Iguazú Falls, Misiones, Argentina, and the other from a vertical cliff permanently swept by a small waterfall at the Brazilian side (the Iguazú River is in the northeastern part of Argentina, and it constitutes the natural geographical border with Brazil at that site). Two other samples were collected from Salto San Martín at the Iguazú Falls and from a small waterfall at the Arrechea Rivulet, the latter 2 km away from the type locality.

The specimens were fixed in 10% formalin after having been partly relaxed by immersion in warm water (55°C) for a few minutes. After relaxation and fixation in formalin, the specimens used for anatomical studies were treated and preserved in a modified Raillet-Henry's solution (distilled water 93%, acetic acid 2%, formalin 5%). The radulae were dissected from the buccal mass, then treated with 10% sodium hydroxide (12 hrs), rinsed in distilled water and properly mounted and coated with gold for scanning electronic microscopic observation.

The type specimen of *Chilina megastoma*, deposited at the Museo de la Plata (MLP, without registration number), has also been studied. Specimens of *C. fluminea* used for comparative purposes were collected from the sandy shore at the Río de La Plata River and from Miguelín Rivulet, both at Punta Lara, Ensenada, Buenos Aires.

Shell measurements (total shell length, last whorl length, major and minor diameter of the shell aperture) were taken under a stereoscopic microscope provided with a micrometer eyepiece.

Voucher specimens were deposited at the malacological collection at Museo de La Plata (MLP nos. 5098, 5099, 5128, 5129, 5130, 5246, and 5261).

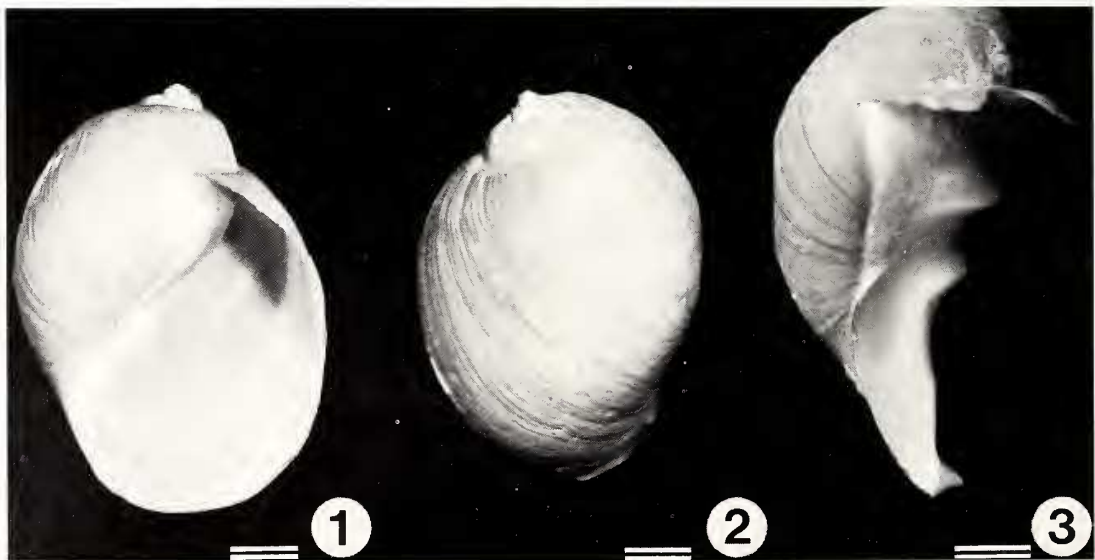
RESULTS

SHELL: The diagnostic features of *Chilina megastoma* are shape and development of the spire (extremely low) and the great development of the last whorl of the shell, resulting in a large aperture of the shell (Fig. 1). The

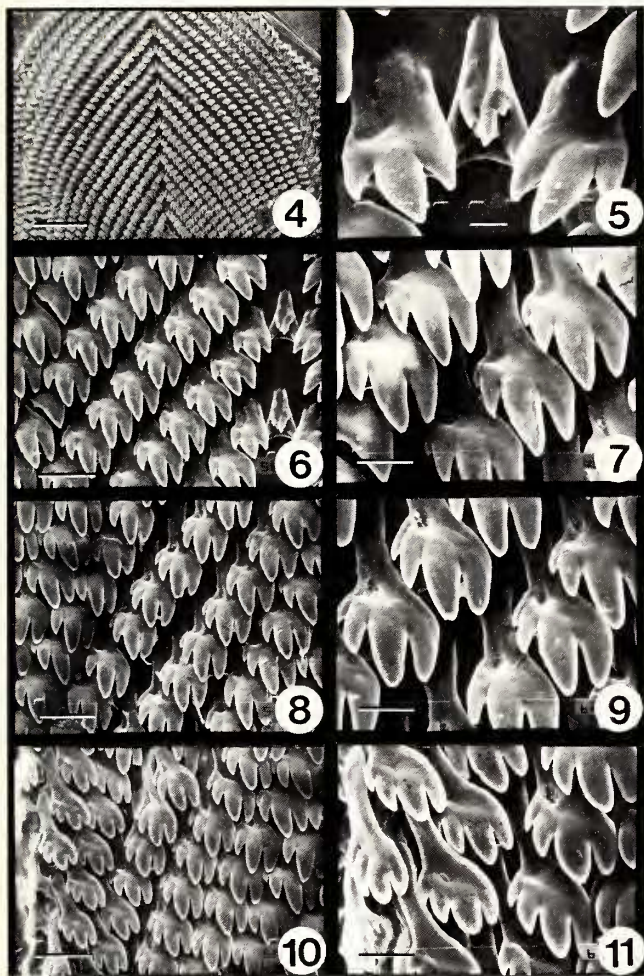
ratio of last whorl to shell length ranged from 0.92-0.97 (mean 0.95, standard deviation 0.01). The aperture is oval; minor diameter to major diameter ratio ranged from 0.53-0.87 (mean 0.67, standard deviation 0.08). The shell is yellowish, olivaceous, or in some cases dark brown. The surface is smooth, only sulcated with weak radial periostracal striae and helicoidal stripes, particularly evident near the suture, which is deeply marked (Fig. 2). The columellar border is straight or slightly curved, somewhat concave, with a marked fold at its upper end (usually referred to as the columellar tooth). The parietal area has a thin white callus. A weak fold of the parietal area forms an indistinct lamella, also referred to in the literature as a "parietal tooth" (Hylton Scott, 1958; Castellanos and Gaillard, 1981), lying slightly over the columellar fold (Fig. 3). The outer lip is sharp and evenly curved.

DIGESTIVE SYSTEM: The general morphology of the digestive system follows the pattern already known in the genus (Haeckel, 1911; Harry, 1964). The posterior end of the radular sac exceeds the posterior end of the buccal bulb, forming a short tube slightly bent dorsally. The salivary glands are not fused at their posterior end.

The radula is composed of approximately 40 sharply oblique rows of teeth (Fig. 4), that is, the tooth rows are arranged "in chevron." The angle between the right and left halves of each row is about 90°. The central tooth is asymmetrical, bicuspid, with an additional third cusp, greatly reduced, and partly overlapping the major cusp (Fig. 5). The attachment base of the central tooth is elongated, deltoid in shape, with the proximal end concave.



Figs. 1-3. Shell of *Chilina megastoma* Hylton Scott, 1958. Fig. 1. Apertural view (MLP 5098). Fig. 2. Dorsal view, showing the periostracal sculpture (MLP 5098). Fig. 3. Detail of the columellar area, columellar fold, and parietal lamella (MLP 5099). Scale bars = 2 mm.



Figs. 4-11. Radula of *Chilina megastoma* Hylton Scott, 1958. Fig. 4. General view showing the typical arrangement of teeth. Fig. 5. Central tooth and first lateral teeth. Figs. 6, 8, 10. Lateral teeth (from central toward marginals). Figs. 7, 9, 11. Details for Figs. 6, 8, 10, respectively. Scale bars = 10 μ m (Fig. 5), 25 μ m (Figs. 7, 9, 11), 50 μ m (Figs. 6, 8, 10), 250 μ m (Fig. 4).

The major cusp is relatively short, cylindrical, and pointed at the tip. The minor cusps are very short and thorn-like. The first lateral tooth is tricuspid, with a short, wide, and strong base. The mesocone has a short dagger-like shape, somewhat directed toward the central tooth. The entocone and ectocone are short, triangular, and nearly symmetrical. The entocone is somewhat larger than the ectocone (Figs. 5-6). The second lateral tooth is also tricuspid, but with a slender, longer, and slightly asymmetric attachment base. The mesocone is the largest cusp, and the ectocone markedly smaller than the entocone (Fig. 6). From the third lateral tooth to the marginal teeth, the morphology of the teeth is relatively similar. The free plate of the teeth is palmate with four cusps; the attachment base is slender, and it joins the free plate at a point slightly shifted toward the minor cusps.

In the last marginal teeth, the attachment base becomes slender, obliquely inserted, and slightly bent (Figs. 7-11).

REPRODUCTIVE SYSTEM: Regarding the general aspects, the features of the female and male genital systems are coincident with those previously described for the genus (Haeckel, 1911; Duncan, 1960a, b, 1975). Thus, only the diagnostic characteristics for *Chilina megastoma* will be described.

Female genital system: The seminal receptacle or spermatheca is oval, located at the left side of the visceral mass, lying just below the ventricle. A long duct runs transversely across the visceral mass, passing over the utero-vaginal complex, to connect the seminal receptacle with the distal end of the vagina just before the female genital opening at the right side of the body (Figs. 12-13). Another duct arises posteriorly, just between the vagina and the uterus,

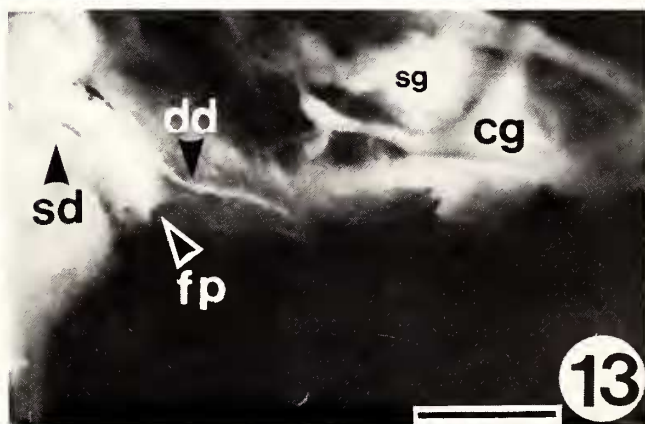
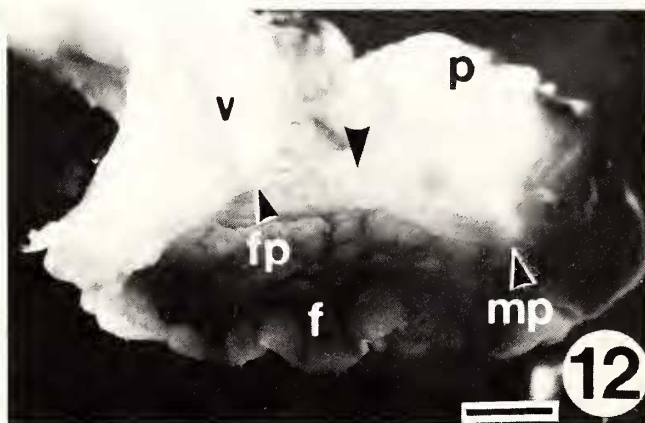
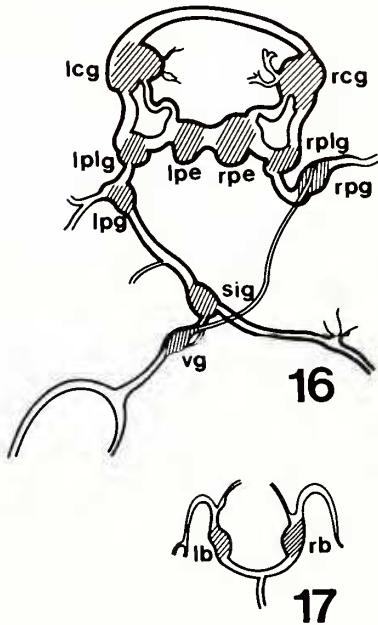
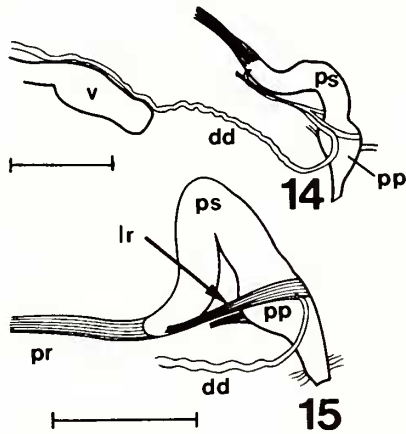


Fig. 12-13. *Chilina megastoma* Hylton Scott, 1958. Fig. 12. Lateral view of a partially dissected specimen showing the terminal portion of male and female genital systems, and the deferent duct (arrow heads) dissected from the body wall (MLP 5210). Fig. 13. Details of the terminal female genital system showing the spermathecal duct crossing over the vagina to open at its distal end (MLP 5210). (cg, cerebral ganglion; dd, deferent duct; f, foot; fp, female pore; mp, male pore; p, penis; sd, spermathecal duct; sg, salivary gland; v, vagina).

called a secondary or accessory seminal receptacle. This duct runs closely adhered to the wall of the uterus, and it is barely enlarged at the tip to form a nearly indistinguishable, rounded ampulla.

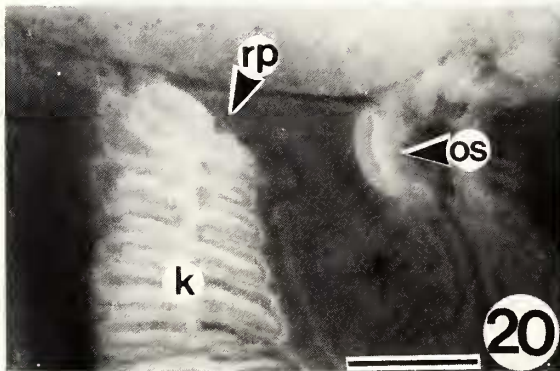
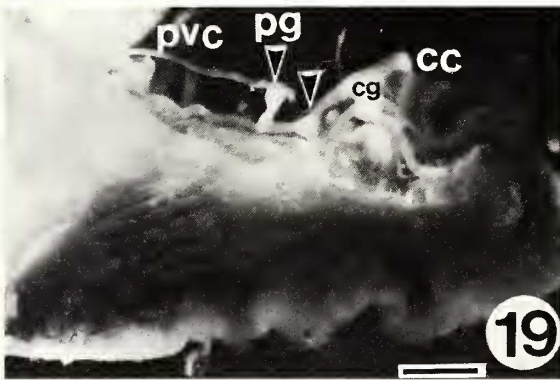
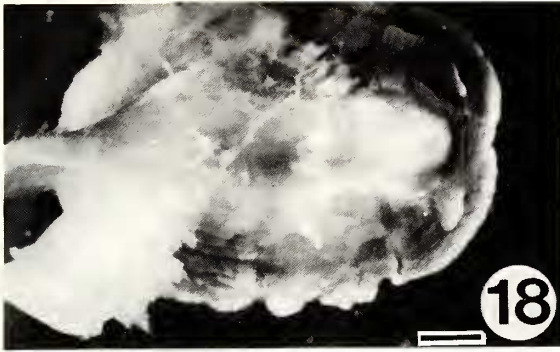
Male genital system: The deferent duct arises as a separate male genital duct only after traversing the prostate,

running below the uterus and the vagina. Near the distal end of the vagina the deferent duct emerges from the haemocoelic space, sometimes forming a very small loop, entering into the muscular body wall (Figs. 12-13). The distal portion of the deferent duct is very sinuous running along the right side of the body wall (Figs. 12-14), close to the surface and immediately below the external reproductive groove which, arising just at side of the female genital pore, runs toward the base of the right tentacle close to the male genital system opening. At this point, the deferent duct diverges from the body wall coming into the cephalo-pedal haemocoel and turning posteriorly toward the posterior end of the penis sheath, the site at which it enters the penis (Fig. 15). The penis sheath is a slender and nearly cylindrical tube (Fig. 14). The penis is slender, as long as the penis sheath; its surface is rugose due to the presence of deep transverse furrows and padded ridges or short lamellae. At the anterior half of the penis, these ridges are more or less longitudinally arranged. The prepuce is a somewhat triangular or cordiform structure with thick muscular walls (Figs. 14-15). The prepuce is shorter than the penis sheath; its length is half or a third of that of the penis sheath. A powerful penis sheath retractor muscle arises from the posterior end of the penis sheath (Fig. 15), and it is attached at the columellar muscle. Another series of four or five muscular bundles are attached to the lateral wall of the prepuce. Two long muscular bundles are joined to the frontal wall of the prepuce running on each side, and connecting with the penis sheath retractor. The contraction of these two muscles determines the erection of the penial complex (Figs. 14-15). The deferent duct runs over the right-most muscular bundle, toward the proximal end of the penis (Figs. 14-15).



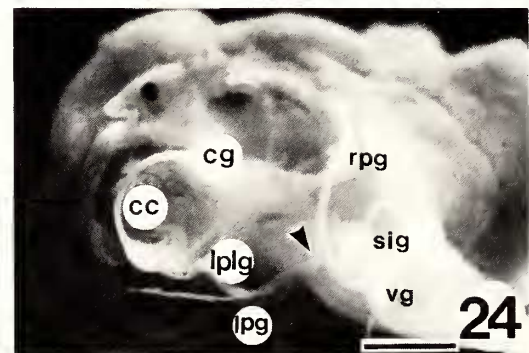
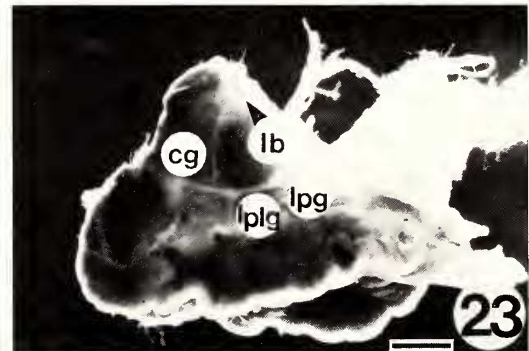
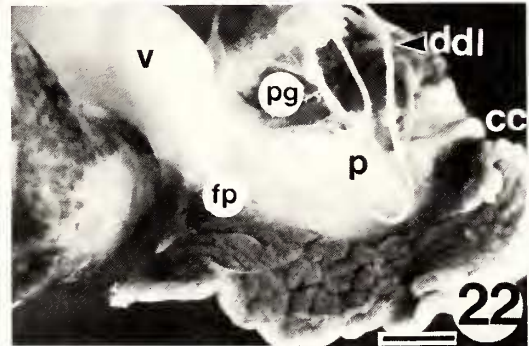
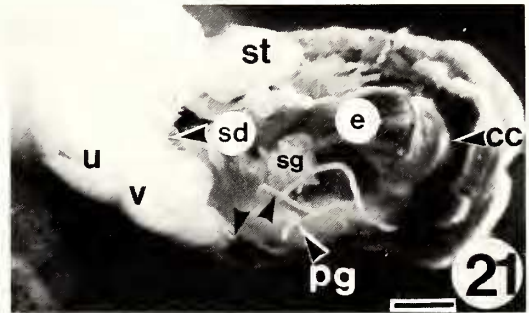
Figs. 14-17. *Chilina megastoma* Hylton Scott, 1958. Genital and nervous systems. Fig. 14. Camera lucida drawing of the distal portion of the female and male genital systems. Fig. 15. Camera lucida drawing with details of the penial complex. Fig. 16. Semi-diagrammatic scheme of the anterior and posterior nerve rings. Fig. 17. Semi-diagrammatic scheme of the innervation of the buccal mass. Scale bars = 1 mm. (dd, deferent duct; lb, left buccal ganglion; lcg, left cerebral ganglion; lpg, left parietal ganglion; lplg, left pleural ganglion; lr, lateral penis retractor; pp, prepuce; pr, penis retractor; ps, penis sheath; rb, right buccal ganglion; reg, right cerebral ganglion; rpe, right pedal ganglion; rpg, right parietal ganglion; rplg, right pleural ganglion; sig, subintestinal ganglion; v, vagina; vg, visceral ganglion).

NERVOUS SYSTEM: The cerebral and pedal ganglia are joined by commissures and connectives to form the anterior nerve ring located at the anterior half of the buccal mass, just at or a little behind the origin of the esophagus (Figs. 16, 18-19). However, the location of the anterior nerve ring showed considerable variation according to the degree of retraction of the buccal mass, but it was always located at the anterior half of the buccal mass. The right pleural ganglion is joined by somewhat short connectives to both cerebral and pedal ganglia. A relatively long connective joins the right pleural ganglion to the right parietal ganglion (supraesophageal ganglion of Fretter, 1975), which gives off a large nerve that supplies the osphradium (Fig. 19), and a second very long and thin connective that runs to the visceral ganglion (also called abdominal [Fretter, 1975] or innominate [Harry, 1964]), at the end of the posterior nerve ring. On the left side of the posterior nerve ring, two short connectives join the left cerebral and pedal ganglia with the pleural ganglion, from which a relatively short



Figs. 18-20. *Chilina megastoma* Hylton Scott, 1958 (MLP 5210). Fig. 18. Dorsal view of a dissected specimen to show the anterior and posterior nerve rings (see Fig. 16 for reference). Fig. 19. Lateral view showing the location of the cerebral commissure. Posteriorly, the cerebral ganglion, the right pleural ganglion (arrowhead), the right parietal ganglion and the parieto-visceral connective, are shown. Fig. 20. View of the mantle roof showing the location of the osphradium close to the renal pore at the right end of kidney. Scale bars = 1 mm. (cc, cerebral commissure; cg, cerebral ganglion; k, kidney; os, osphradium; pg, parietal ganglion; pvc, parieto-visceral connective; rp, renal pore).

connective arises and runs toward the left parietal ganglion. A long connective joins the left parietal ganglion to the subintestinal ganglion, which lies over the columellar muscle, at its posterior half. A somewhat large nerve arises at a point approximately 2/3 the length of the left parietal-subintestinal connective. There is a slight swelling just at the site where that nerve originates; however, it might not



Figs. 21-24. *Chilina fluminea* (Maton, 1809) (MLP 5246). Fig. 21. Dorsal view of a partially dissected specimen showing position of the cerebral commissure, and the long loops of the deferent duct (arrowheads). Fig. 22. Right lateral view. Fig. 23. Left lateral view. Fig. 24. Dorsal view of the nervous system. Scale bars = 1 mm. (cc, cerebral commissure; cg, cerebral ganglion; ddl, deferent duct loop; e, esophagus; fp, female pore; lb, left buccal ganglion; lpg, left parietal ganglion; lplg, left pleural ganglion; p, penis; pg, parietal ganglion; rpg, right parietal ganglion; sd, spermathecal duct; sg, salivary gland; sig, subintestinal ganglion; st, spermatheca; u, uterus; v, vagina; vg, visceral ganglion).

definitely be considered a ganglion. Finally, a very short connective joins the subintestinal ganglion to the visceral one, closing the posterior nerve ring. From the latter, a large nerve arises, and shortly thereafter, it gives off two nerves which supply the visceral mass. From the subintestinal ganglion, a large nerve runs toward the right, passing through the columellar muscle to innervate the distal part of the vagina and the pneumostomal appendage. As is characteristic for the genus, the pleuro-visceral connectives show incomplete torsion (Figs. 16, 18).

At each side of the buccal mass (dorsolaterally), far removed, and behind the origin of the esophagus, are the buccal ganglia. The ganglia are joined by a somewhat short commissure which passes below the esophagus. A nerve, arising from the middle of this commissure, sinks into the buccal mass shortly after its origin (Fig. 17). The buccal ganglia are connected to each cerebral ganglion by long connectives, forming an U-shaped open nerve ring. The osphradium, located on the roof of the mantle cavity, close to the renal pore, and at the anterior end of the pneumostome, is a flat oval-shaped organ formed by an slightly curved furrow, bordered by two elongated and inflated lips (Fig. 20).

DISCUSSION

From the present knowledge of the distribution of *Chilina megastoma*, the species seems to be endemic to a reduced number of environments closely related to high energy freshwater courses such as vertical cliffs and rocky walls permanently swept by winds and water trickles from nearby waterfalls. As reported for other gastropod species (Trussell *et al.*, 1993), the extreme reduction of the spire, the globose shape of the last whorl and the wide aperture of the shell shown by *C. megastoma* represent adaptive responses to particular environmental conditions, such as dislodgement agents in high energy watercourses.

Hylton Scott (1958), and Castellanos and Gaillard (1981) considered that *Chilina megastoma* approaches *C. fluminea* in shell morphology, pointing to the lack of periostracal sculpture as a differential character in the latter.

With respect to the anatomy of soft parts, several differences should be pointed out. The general features of the genital system of *Chilina megastoma* are coincident with those described by Miquel (1984) for *C. fluminea* and by Harry (1964) for *C. fluctuosa* Gray, 1828. The following differences have been observed: in *C. megastoma* the duct of the seminal receptacle or spermatheca crosses, from left to right, above the lower portion of the female genital complex, just below the floor of the pallial cavity, while in *C. fluminea*, the duct passes over the anterior border of the distal part of the spermooviduct and prostate, and then turns

downward, just at the point where the uterus connects with the vagina (Fig. 21). Thereafter, to traverse the vagina ventrally, the duct reappears dorsally and immediately opens near the tip of the vagina. The same features (Fig. 16) have been illustrated by Harry (1964) for *C. fluctuosa* from Chile. Moreover, in *C. megastoma*, the accessory or secondary seminal receptacle is barely enlarged at the tip, while in *C. fluminea* the tip of this organ is greatly enlarged into a large pear-shaped bulb. This difference in shape and size does not seem to be related to organ physiology.

In the male portion of the genital system, several differences among species should be pointed out: while in *Chilina megastoma* the segment of the deferent duct which runs into the muscular body wall just below the genital groove is markedly sinuous (Figs. 12, 14), in *C. fluminea* it is nearly always straight or faintly sinuous (Fig. 22). In addition, in *C. fluminea*, when the deferent duct reenters into the haemocoelic space toward the penis, it develops two or three large loops prior to running along the penial retractor muscle and entering the penis (Figs. 21-22). Unlike this, in *C. megastoma* the deferent duct is strictly as long as the genital organs it accompanies (Figs. 12, 14-15).

According to Valdovinos and Stuardo (1995), the nervous system is the only reliable feature to differentiate Chilean species and to build suitable supraspecific arrangements. According to Haeckel (1911), the structural pattern of the posterior nerve ring seems to be a specific characteristic, especially with respect to the number of ganglia and the length of the connectives between the left pleural and subintestinal ganglia.

The general pattern of the nervous system of *Chilina megastoma*, here studied, does not differ from that described by Harry (1964) for *C. fluctuosa*. However, Harry (1964) was unable to find the left parietal ganglion already described by Haeckel (1911). In the present study, the left parietal ganglion was easily identified in both *C. megastoma* and *C. fluminea* (Figs. 18, 23-24). In *C. megastoma*, the lengths of the left cerebropleural connective and left pleuroparietal connectives were similar. The left parietal-subintestinal connective was approximately three times as long the pleuroparietal connective (Fig. 18). In *C. fluminea* the left pleural and parietal ganglia are closer than they are in *C. megastoma*, being, in some cases, nearly contiguous (Fig. 24). In *C. megastoma* the anterior nerve ring tends to be located more posteriorly than in *C. fluminea*, in which it has always been observed at the anterior end of the buccal bulb (Figs. 21-23).

As in *Chilina fluminea*, the posterior end of the radular sac of *C. megastoma* is projected from the buccal mass, forming a short cylindrical tube bent slightly dorsally. This fact is in contrast with the description given by Harry (1964) for *C. fluctuosa* from Chile.

Several details in morphology of the radular teeth



Figs. 25-26. Radula of *Chilina fluminea* (Maton, 1809). Fig. 25. Details of central and first lateral teeth. Fig. 26. Lateral teeth. Scale bars = 25 μ m (Fig. 25), 50 μ m (Fig. 26).

seem to differ consistently among species. In *C. megastoma*, the central tooth has a definite triangular, elongated base, while in *C. fluminea* the attachment base is shorter and stronger. The lateral and marginal teeth of *C. megastoma*, have more slender attachment bases and their cusps are longer, with a more definite dagger-like shape than those of in *C. fluminea* (Figs. 25-26). In addition, the last marginal tooth of each row in *C. megastoma* has four cusps (Figs. 10-11), while in *C. fluminea* it usually has six or seven cusps (Castellanos and Gaillard, 1981). The number of tooth rows also differs: 40 in *C. megastoma* and between 60 and 65 in *C. fluminea* (Castellanos and Gaillard, 1981).

LITERATURE CITED

- Castellanos, Z. J. A. and M. C. Gaillard. 1981. Mollusca, Gasteropoda, Chiliniidae. *Fauna de Agua Dulce de la República Argentina* 15(4):20-51.
- Castellanos, Z. J. A. and N. A. Landoni. 1995. Mollusca Pelecypoda y Gastropoda. In: *Ecosistemas de Aguas Continentales, Metodologías Para su Estudio, T. 2*, E. C. Lopretto and G. Tell, eds. pp. 759-801. Ediciones Sur, La Plata, Argentina.
- Castellanos, Z. J. A. and S. E. Miquel. 1980. Notas complementarias al género *Chilina* Gray (Mollusca, Pulmonata). *Neotropica* 26(76):171-178.
- Castellanos, Z. J. A. and S. E. Miquel. 1991. Distribución de los Pulmonata Basommatophora. *Fauna de Agua Dulce de la República Argentina* 15(9):1-11.
- Duncan, C. J. 1960a. The evolution of the pulmonate genital system. *Proceedings of the Zoological Society of London* 134:601-609.
- Duncan, C. J. 1960b. The genital systems of the freshwater Basommatophora. *Proceedings of the Zoological Society of London* 135:339-355.
- Duncan, C. J. 1975. Reproduction. In: *Pulmonates, Vol. I. Functional Anatomy and Physiology*, V. Fretter and J. Peake, eds. pp. 309-365. Academic Press, London.
- Fretter, V. 1975. Introduction. In: *Pulmonates, Vol. I. Functional Anatomy and Physiology*, V. Fretter and J. Peake, eds. pp. xi-xxix. Academic Press, London.
- Harry, W. H. 1964. The anatomy of *Chilina fluctuosa* Gray reexamined, with prolegomena on the phylogeny of the higher limnic Basommatophora (Gastropoda: Pulmonata). *Malacologia* 1(3):355-385.
- Haeckel, W. 1911. Beiträge zur Anatomie der Gattung *Chilina*. *Zoologische Jahrbücher*, supplement, 13(4):89-136.
- Hylton Scott, M. I. 1958. Nueva especie de *Chilina* del norte argentino (Moll. Pulm. Basommatophora). *Neotropica* 4(13):26-27.
- Miquel, S. E. 1984. *Contribución al Conocimiento Biológico de Gasterópodos Pulmonados del Área Rioplatense, con Especial Referencia a Chilina fluminea* (Maton). Doctoral Thesis, Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata. 133 pp.
- Miquel, S. E. 1986a. El ciclo de vida y la evolución gonadal de *Chilina fluminea fluminea* (Maton, 1809) (Gastropoda Basommatophora Chiliniidae). *Neotropica* 32(87):23-34.
- Miquel, S. E. 1986b. Tipos celulares del tracto genital de *Chilina fluminea fluminea* (Maton, 1809) (Gastropoda Basommatophora Chiliniidae). *Neotropica* 32(88):133-138.
- Trussell, G. C., A. S. Johnson, S. G. Rudolph, and E. S. Gilfillan. 1993. Resistance to dislodgement: habitat and size-specific differences in morphology and tenacity in an intertidal snail. *Marine Ecology Progress Series* 100:135-144.
- Valdovinos, C. and J. Stuardo. 1995. Morfología funcional de *Chilina angusta* (Philippi, 1860), y evolución de Chiliniidae. *Resumos, II Congresso Latino-Americano de Malacologia*, Porto Alegre, Brasil:43.

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